Remarks

1. – 2. Pending Claims/Election/Restriction:

Applicants' Agent acknowledges the withdrawal of Claims 19, 21 -57 and 64 -94 as being drawn to non-elected inventions, including the Examiner's maintenance of the requirement for the election of "soil sample" as a species, resulting in the withdrawal of claims 19, 21 and 22 from further consideration. Applicants' Agent has therefore herein cancelled Claims 19, 21-57 and 64-94. In addition, the Agent for the Applicants has cancelled Claims 58 – 63. Therefore, the Claims remaining include Claims of Group I, Claims 1 – 18, 20, as originally filed and new Claim 95. New Claim 95 contains no new matter.

5. – 6. Priority:

The Examiner states:

"The disclosure of the prior-filed application, Application No. 10/113,916, fails to provide adequate support or enablement in the manner provided by the first paragraph o 35 U.S.C. 112 for one or more claims of this application. Application No. 10/113,916 does not appear to provide support for an invention that is a method for analyzing the organismic complexity of a sample, and comprising relating the listing of signature tags to DNA sequences in database to determine the variety and relative numbers of organisms originally present in the sample, as in claim 1. Therefore, the invention of the claims examined herein, is afforded priority to the filing date of instant application, i.e., 3/2/2004."

The Agent for the Applicants respectfully disagrees and requests reconsideration of the priority of the invention of Claim 1 as being the date of filing of the original priority document, i.e., U.S. Application No. 10/113,916, filed April 1, 2002. The Agent requests reconsideration on the basis of the following remarks as well as remarks included in response to items 13 and 14 of the Office Action:

The method of Claim 1 is fully supported by the priority document, U.S. Application No. 10/113,916, filed April 1, 2002. The method claimed in the priority document contains all of the steps necessary to create a "genome signature tag library". And, the specification of the priority document clearly supports the relating of the genome signature tag library to databases to determine the "DNA source" (i.e., the organisms that contributed DNA to the sample) and to obtain an estimate of the relative numbers of different organisms as shown by the following statements of the priority application document:

The statement on page 2, line 25 through page 3, line 3, as follows, shows that the invention, fully described in the priority application, is directed to a method for the identification and quantitation of microorganisms isolated from natural habitats:

"Seeking a comprehensive DNA-based **method for the identification and quantitation of microorganisms isolated from natural habitats**, a RLGS approach with Serial Analyses of Gene Expression (SAGE) principles **were combined to create an open architecture technique that provides Genomic Signature Tags (GSTs)**." (emphasis added)

On page 4, lines 12 to 18, the statement:

"This length suggested that MmeI could be used to obtain unique tags directly from total microbial DNA since there are 4^{21} or more than 4 trillion possible 21-mer tag sequences, which by far exceeds the number of 21-mers in most microbial genomes. Consequently, **a MmeI tag should, in most cases, be able to uniquely identify its DNA source even in the absence of positional information**."

clearly indicates that the tags generated by the disclosed technique are adequate and useful for identifying the source DNA, i.e., the organism(s) comprising the organisms of a sample.

On page 4, line 27 through page 5, line 2, the statement:

"The resulting **sequences are identified through database matches** or used to create a new database that is specific for a particular DNA sample."

clearly shows that the sequences are to be related to (identified through) sequences in databases in order to identify (as in lines 12 to 18) "its DNA source" – i.e., the organism(s) from which the GST tag originated.

Combining the statement from pages 4 to 5, noted immediately above, with the statement from page 10, line 25, through page 11, line 4 indicates that one can identify the DNA source "**if the sequence is available**" – in other words, if the sequences obtained from a sample are already available in DNA sequence listings in databases one can identify the organism (DNA source) possessing that sequence and therefore identify the organism(s) in the sample:

"The results of this study show that the GST technique provides a route to obtaining numerous 21-22 bp sequence tags that can be used to identify the DNA source, and as shown here, the presence or absence of particular tags can provide some indication of the genetic variability between two closely related strains. The length of the tags allow **direct determination of the source DNA if the sequence is available**."

The specification of the priority application goes on then to note that with entire genome sequences that are already present in databases for sixty (60) micro-organisms, the identification of individual species (and even sub-strains of species) is possible because there are very few shared GSTs among the organisms and many GSTs that are unique to each species (or sub-strain). See lines 4 – 15 of page 11 of the priority application, as follows:

"In silico comparison of all the BamHI-NlaIII GSTs that would be generated from a mixture of the 60 complete microbial genomes,

the NCBI database demonstrated that these different bacterial strains share few GSTs in common. Table 4 contains a list of the top 30 shared tags. The worst case scenario is the occurrence of a single tag that was found three times in E. coli and once in Y. pestis. No GST was shared by three strains, although this might change as more closely related organisms are sequenced. Even between closely related strains, the frequency of unique, unshared identifiers is more than adequate to allow strain differentiation."

In the above quotation, one should also take note of the penultimate sentence:

"No GST was shared by three strains, although this might change as more closely related organisms are sequenced."

since the statement infers that "as more closely related organisms are sequence . . ." and the new sequences are added to databases more sequences will be available to relate to GSTs found in a sample.

In lines 16 to 19 of page 13 it is clear that the method of the priority document can be used to identify the organisms comprising a sample and to provide a means for relative quantification of the organisms of the sample:

"This means that the technique should, in addition to being able to identify DNA sources, be able to provide a fairly accurate means for quantitative analysis of mixed DNA populations."

And, in summary, from page 15, line 10 to 13:

"In summary, the basic GST procedure provides a means for genome-wide fingerprinting of chromosomal and episomal DNAs, and by extension, for compositional analysis of natural populations."

And, from the Abstract, lines 13 to 15:

"The tag sequences and abundances are used to create a GST profile that can identify and quantify the genome of origin within any complex DNA isolate."

In conclusion, the Agent for the Applicants submits that the priority application, U.S. application No. 10/113,916:

1. **does provide** support for an invention that is a method for analyzing the organismic complexity of a sample; and

2. **does comprise** relating the listing of signature tags to DNA sequences in database(s) to determine the variety and relative numbers of organisms originally present in the sample.

Therefore, the Agent respectfully requests priority designation of US Patent Application No. 10/133,916, filed April 1, 2002.

Thus, the Agent respectfully requests reinstatement of the priority date as April 1, 2002.

7. Information Disclosure Statement

Notation acknowledged

8. Specification

The amendment to the specification suggested by the Examiner has been added on page 2 of this paper. The amendment of the specification contains no new matter.

9. – 10. Claim Rejections – 35 USC § 112 – First Paragraph, Written Description

The Examiner indicates a rejection of all claims currently pending for failing to comply with the written description requirement of 35 USC 112, first paragraph.

The Examiner cites the publication of van der Lelie, et al.: Applied and Environmental Microbiology (March, 2006, Vol **72**, pp 2092-2101), as suggesting that the written description of the GST method found in the present specification fails to comply with the written description requirements of 35USC 112. The Agent for the Applicants respectfully disagrees and requests that the Examiner reconsider and withdraw this rejection because the cited publication, and specifically the specific citations quoted by the Examiner, fully and completely show that the specification of the present application fully complies with the 35USC 112, paragraph 1 written description requirements.

The specific citation of the Examiner as follows:

"most environmental communities are far too complex to be fully sequenced using metagenome shotgun sequencing."

indicates that something **other than** shotgun sequencing of a community is required and does NOT suggest that the GST methods would be unsuitable – in fact, quite the contrary, the statement suggests that GST methods would be suitable. In fact this statement serves to justify the development of the GST method, and its sub-method, the "Single-Point GST" methods of the cited article.

And, further:

"in many cases **individual** GST sequences provided **sufficient specificity for species identification**"

indicates that the GST methods are **sufficient to identify species of a community!** To be specific, the latter quotation from the cited article indicates that only a few **individual GST sequences** are needed in order to identify any given organism present in a community.

Therefore (as per the cited article) "This result prompted us to look for fragmenting enzymes that

would generate **only one or a few *informative* tags per organism**." In other words, the object of the "Single-Point" GST cited article (and claimed as Group III (now withdrawn and cancelled) in the present application) was to reduce the number of GSTs that would be generated from any given sample because "**individual GST sequences provided sufficient specificity for species identification**". Thus, the cited article, rather than indicating that the GST method of the present invention is not supported by an adequate written description does in fact **fully** support quite the direct opposite, i.e., that the methods described in the present specification and claimed in the pending claims is fully described.

For the above reasons, therefore, the Agent respectfully requests withdrawal of the rejection based upon 35 USC 112, first paragraph written description.

11. Claim Rejections – 35 USC § 112 – First Paragraph, Enablement

The Examiner indicates a rejection of all claims currently pending because the "specification **while being enabling for identifying and analyzing closely related species or strains and simple, defined microbial communities,** does not reasonably provide enablement for analyzing the organismic complexity or any sample, including diverse, complex environmental samples. The specification does not enable any person skilled in the art . . . . to make and use the invention commensurate in scope with these claims." The Examiner then cites the Wands factors.

Again, the Agent for the Applicants respectfully disagrees and requests the Examiner's consideration of the following remarks, and in view thereof, reconsideration and withdrawal of this rejection.

1 and 2; 3 and 5: The breadth of the claims and the nature of the invention; the state of the prior art and level of predictability in the art:

Again the Examiner cites the van der Lelie, et al. Applied and Environmental Microbiology article implying that the statement therein: "in many cases the individual GST sequences provided sufficient specificity for species identification" suggests that the GST method of the present claims is not enabled by the present specification. As above, this statement has been misinterpreted by the Examiner and the statement, in fact, fully indicates that the GST method of the present invention is enabled. The statement means that because "**the individual GST sequences** provided sufficient specificity for species identification", the entire spectrum of GST sequences DOES NOT need to be analyzed in order to identify species in a community, indicating that the GST method is fully enabled as a method for determining the organismic complexity of a sample.

From the same reference the Examiner then cites:

"As has been documented for other PCR-based methods, amplification biases lead to misrepresentation of the overall community composition. It was concluded that the great strength in this technology lies in its discriminatory power."

as suggesting that the GST method is like PCR-based methods wherein amplification biases lead to misrepresentation. On the contrary, the second sentence of the Examiner's citation is clearly directed at supporting the "great strength" of the GST method and its sub-method, the "Single-Point GST" method. The last quoted sentence:

"Given its open architecture, diverse application, and the facility with which we can link tags to any gene of interest, the use of SP-GSTs has great potential and application for identifying and analyzing closely related species or strains and simple microbial communities."

speaks solely to the sub-method, i.e. the Single-Point GST method and not to the overall GST method as covered in the present claims. This statement, taken together with the statement "**the individual GST sequences** provided sufficient specificity for species identification" indicates that the GST method is superior to PCR-based methods wherein amplification biases lead to a misrepresentation of the overall community composition.

4. The relative skill of those in the art.

The Examiner states that the level of skill in the art would be **"high, most likely at the Ph.D. level"** but that such persons would be required to engage in undue experimentation to carry out the invention due to its unpredictability.

The Agent for the Applicants wishes to point out the following statement from the specification, paragraph [0051], second sentence:

"The method can be performed with equipment available in most molecular biology laboratories."

Ordinary molecular biology laboratory equipment (and skill level) is all that is required. Each of the steps of the GST method is understood (and readily practiced) by persons of ordinary skill in the art, and would be well understood and expertly practiced by those of "high, most likely Ph.D. level" skill. Thus, there would be no "undue experimentation" required to carry out the invention, but one would merely have to practice the steps in the order called for in the claims.

6-7. Amount of direction provided and the existence of working examples.

The Examiner indicates an expectation that more examples would be required for a practitioner to make use of the invention as disclosed in the specification and as claimed in the pending claims.

The Agent respectfully points out the MPEP 2164.02 states: "Compliance with the enablement requirement of 35 USC 112, first paragraph, does **not** turn on whether an example is disclosed." And, "The specification need not contain an example if the invention is otherwise disclosed in such manner that one skilled in the art will be able to practice it without an undue amount of experimentation". Further: ". . . lack of working examples or lack of evidence that the claimed invention works as described should never be the sole reason for rejecting the claimed invention on the grounds of lack of enablement." And "For a claimed genus, representative examples together with a statement applicable to the genus as a whole will ordinarily be sufficient if one skilled in the art (in view of level of skill, state of the art and the information in the specification) would expect the claimed genus could be used in that manner without undue experimentation. Proof of enablement will be required for other members of the claimed genus only where adequate reasons are advanced by the examiner to establish that a person skilled in the art could not use the genus as a whole without undue experimentation."

The Agent for the Applicant wishes for the Examiner to consider whether he has established adequate reasons why a person of skill could not use the genus as a whole without undue experimentation. The citation of the Examiner: "one must develop various strategies for assuring that a statistically significant number of tags have been sequenced" does not establish that one of skill would be required to carry out "undue experimentation". Further stating that the specification "contemplates various possible strategies for addressing these limitations" indicates that the specification is enabling for those of skill in the art by having made those of skill aware of those strategies.

The Agent is unable to determine how the statement "use of a rare-cutting fragmenting enzyme will not always serve the purpose of reducing the number of GSTs generated from

organisms and would certainly increase the likelihood of failing to detect some of the organisms comprising a sample" relates to enablement of the claims pending in this case since the statement in the specification is made as a reason to have contemplated (and developed ("enabled")) the Single-Point GST method.

Given the amount of description of various alternatives in practicing the claimed invention it is the position of the Agent for the Applicants that those of skill in the art could readily practice the invention, using ordinary molecular biology laboratory equipment, without any experimentation that would be considered by them as undue. The practitioners of this art are accustomed to making choices as to fragmenting enzymes, binding pairs, type IIS enzymes, methods of sequencing, and etc. based upon their experience, their review of information available to them in databases and the literature, and etc. None of such practitioners would consider making such choices as being "undue". None of the practitioners would consider it unreasonable to make such choices and would find the guidance and, particularly the contemplation of "various possible strategies for addressing these limitations" that are found in the specification, to make such choices easier to make.

8. Quantity of experimentation to make or use the invention.

The Agent refers the Examiner to the above remarks. In addition, the Agent wishes to suggest to the Examiner that the only "unpredictable" aspect of the invention would be the actual outcome when a practitioner begins a study of a new sample – practicing the methods of the invention claimed here **will always generate** a result regardless of what the sample is, provided the sample contains one or more organisms as required in step a) of Claim 1. Once the new sample is analyzed the practitioner can then make a "skilled" choice as to whether some of the

"various possible strategies" presented in the specification may or may not provide additional useful information.

In summary, for the above reasons, therefore, the Agent respectfully requests withdrawal of the rejection based upon 35 USC 112, first paragraph, enablement.

12. Claim Rejections – 35 USC § 103

On page 12 of the Office Action, the Examiner has noted the naming of joint inventors. The Agent for the Applicants affirms that the subject matter of the various claims was commonly owned at the time the covered inventions were made.

13. Claim Rejections – 35 USC § 103(a) – Zhou et al. (J. App. Micro. 2001 **90**:96) in view of Dunn et al. (Genome Res. 2002 **12**:1756) and Zhou et al. (J. App. Micro. 2001 **90**:96) in view of Dunn et al., US 2003/0186251 A1 (published 10/2/2003)

The Examiner has indicated that the invention would have been obvious to one of skill in the art in view of the teachings of Zhou et al. in combination with those of Dunn et al.. The Examiner states:

"Dunn et al., (11/2002), Genome Research, Vol. 12, pp1756-1765 . . . **teach a method for analyzing the organismic complexity of a sample**" and Dunn et al., US 2003/0186251 A1 (published 10/2/2003) . . . **teach a method for analyzing the organismic complexity of a sample**."

First, the Agent wishes to draw the Examiner's attention to the fact that the text and figures 1 and 2 and all the Tables of the Dunn et al. (11/2002) Genome Research reference and the specification and tables and the Drawings of the Dunn et al. US 2003/0186251 A1

publication are **virtually identical** although arranged in different orders. Thus, the Dunn et al. Genome Research reference clearly post-dates the US patent application by the inventors.

The Agent respectfully points out that the priority application from which this pending application draws its priority (US Patent Application No. 10/113,916, filed April 1, 2002) clearly pre-dates the Dunn et al. publication (November, 2002). Furthermore, because the full scope of the claims pending in this case are contemplated in the priority patent application and pre-dates the Dunn et al. reference, the Dunn et al. Genome Research reference is not eligible as prior art to be combined with that of Zhou et al. to create the stated case for obviousness.

A similar argument can be drawn regarding the combination of Zhou et al. with the published US Patent Application No. 10/113,916, filed April 1, 2002 (publication number US 2003/0186251. Examination of the priority application and the Dunn et al. reference shows that the basic GST scheme in the reference was Figure 1 of the 10/113,916 application. Furthermore, the Tables 1, 2, 3 and 4 of the Dunn et al. reference were Tables 1, 2, 3 and 4 of the priority application and Figure 4 of the Dunn et al. reference was Figure 2 of the priority application. Thus, all teachings of the Dunn et al. reference cited by the Examiner were contained in the priority US Patent Application No. 10/113,916. In addition, all that the specific methods detailed in the Dunn et al. reference are identical to methods of the Methods in the priority US Patent Application No. 10/113,916.

In addition, as noted above, it occurs to the Agent for the Applicants that **if**, as the Examiner states: "Dunn et al., (11/2002) Genome Research, Vol. 12, pp. 1756-1765 (IDS, entered 7/6/2004) . . . teach a method for analyzing the organismic complexity of a sample", and **if**, as the Examiner states: Dunn et al., US 2003/0186251 A1 (published 10/2/2003) . . . teach a method for analyzing the organismic complexity of a sample" and **if**, as the Agent points out

above, the teachings of Dunn et al. are all contained within the priority application US Patent application number 10/113,916, filed April 1, 2002, then the invention as claimed herein is fully enabled and fully described in the written description of the present application.

Therefore the Agent respectfully requests reconsideration and withdrawal of the rejection based upon the teachings of Zhou et al., in view of both Dunn et al. references

14. Claim Rejections – 35 USC § 103(a): Unpatentable over Zhou et al, J. App. Micro. (2001), Vol. 90 pp96-105; Li et al., (US 2003/0165923 A1 filing date 1/20/2000) (priority to Provisional application No. 60/215,596, 1/20/2000), and Velculescu et al. (US 6,498,013 B2, filing date 7/28/2000) (priority to Provisional application No.s 60/221,556, 7/28/2000 and 60/233,431, 9/18/2000).

The Agent for the Applicants respectfully points out, particularly in view of the remarks under item 13., above, that Li et al., US 2003/0165923 A1 was published on August 7, 2003 and Velculescu et al., (US Application 09/916,228) was publish (US 2003/0008290) on January 9, 2003. Both publication dates are later than the priority date of the claims pending in the present application. Thus neither the Li nor the Velculescu references are eligible as prior art for combination with Zhou to create a case for a rejection of the present claims under 35 USC 103(a).

As stated in item 13., above, and as discussed in great detail in items 5 -6, above, the Examiner indicates that the Dunn et al. references (Genome Research (2002) **12**:1756-1765 and US 2003/0186251 A1) **teach a method for analyzing the organismic complexity of a sample**. Therefore the Agent requests reconsideration and withdrawal of this rejection of the claims pending in this patent application.

Summary

The Agent for the Applicants states that the amendment of the specification and the new Claim 95 contain no new matter. In view of the above amendments and remarks, the Agent for the Applicants respectfully seeks the Examiner's thoughtful review and anticipates a timely issuance of a notice of allowance.

Respectfully submitted,

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